Bacteria Prevalence On The Environmental Labour Wards As The Causes Of Nosocomial Infections In General Hospital Umuguma And Umezuruike Hospital Owerri, In Imo State, Nigeria.

[1]Ijioma B.C Ph.D [2] Kalu I. G. [1]Nwachukwu C.U Ph.D

[1]Department of Biology Alvan Ikoku Federal College of Education, Owerri, Imo State,
[2]Department of Biotechnology Federal University of Technology Owerri Imo State.
nwachukwucu2005@yahoo.co.uk

Abstract: This study was on the isolation and identification of bacteria prevalent on the environmental labour wards of General hospital Umuguma and Umezuruike hospital, Owerri, Imo State of Nigeria. In carrying out the research, samples were collected from the floor, bed rails, walls, mattresses, malting touch and baby beds using sterilized swab stick that is moistened with peptone water. Appropriate media, agar and reagents were used to culture the micro-organism for 24 hours. The organisms were isolated and it was observed that staphylococcus aureus, Bacillus species, pseudomonas acruginosa, Escherichia coli, Streptococcus faecalis, Klebsiella acrogens, and Micrococcus species were predominant in descending order of prevalence. These bacteria are therefore factually suggested to be the major cause of Nosocomial infections in the environmental labour wards of the named hospitals. Recommendations such as keeping hand clean by washing thoroughly with soap and water, or using alcohol base were made for the effective control and reduction of the said infections in the hospitals. [Report and Opinion 2010;2(9):45-52]. (ISSN: 1553-9873).

Key words: Bacteria, Nosocomial, Labour ward, Pseudomonas acruginosa, General hospital

INTRODUCTION

One of the major discoveries in the world of microbiology is the discovery of bacteria in 1683 by a Dutch microscopist and Naturalist, Antonio Van Leewenhock. Bacteria are theref0re described as the microscopic, filamentous, non-green plants, devoid of nuclear membrane. They are mostly saprophytic but often parasitic and usually reproduce by binary fission. (Butani, 2006). Bacteria are classified into four according to their shape as bacilli (rod shaped), cocci (spherical), commas (twisted) and spirilli (spiral). Irrespective of their shape, bacteria are minute and single- celled of about 0.01mm long. (Arora, 2005). Many of these bacteria are economically important as regards their activities in recycling of organic materials by enhancing decay, conversion of atmospheric Nitrogen into Nitrates for plant use, breaking down of food materials in the gut of animals and obviously harmful to organisms by causing diseases (Aroar, 2005).

Microbiological activities in the late 1800s improved the ability to culture and identify bacteria through biochemical tests which are still used till date. (Levy et al; 2001). Paul Ehrlich introduced the method of staining and identifying bacteria and also laid the basis for the development of antibacterial therapy. (Bochud et al; 2001). He hypothesized that "if bacteria could take up dye selectively,, then it might be possible to develop a "magic bullet" to kill bacteria but spare the surrounding cells.

Nosocomial infections (Hospital acquired infections) are referred to as infections that are acquired within the hospital environment between 2-4 days of admission into the hospital or other health care facilities. Nosocomial infections are caused by bacteria, viruses, fungi

or any other parasitic organism. They are contracted by the contaminated equipments of the hospital. Sepsis is a serious rapidly progressive multi-organ infection (blood poisoning) which can lead to death. This could be caused by Nosocomial infections. Nosocomial infections can develop from surgical procedures, insertion of catheters (tubes) into the body, aspirated (inhaled) materials, etc. Nosocomial infection include urinary tract infection, pneumonia, surgical wound infections etc. All patients are at the risk of being infected especially the young, the elderly and persons with compromised immune system. Prolonged hospital stay, severity of the underlying illness, compromised nutritional and immune status, prevalence of antibiotics-resistant bacteria from the over use of antibiotics, failure of health care workers to wash their hands before and after each procedures, etc, are factors that predisposes patients to the Nosocomial infections. However, these factors can be controlled by the use of aprons during patient care, hand washing and gloving, surface sanitation mitigation. (Chan et al, 2007).

Development of additional antibiotics followed the observation of cocci growth on a culture plate in the vicinity of a mould, by Alexander Fleming's in 1928. The coccigrowth was later identified as Penicilium notatum from which Penicillin was isolated. Production concentration and clinical use of penicillin were put forth by Howard Florey and Ernest Chain 1941. In the 19^{th} century, the application of antiseptic techniques becames successful and the "germ theory" was accepted. By the 20^{th} century, the dream of treating infections with antibiotics became a reality. (River et al; 2005). The past decades have witnessed a change in the prevalence of hospital associated bacteria from hemolytic streptococci and penicillin-susceptible staphylococci and relatively susceptible gram negative rod and fungi. Antimicrobial treatment alters the normal flora, facilitates colonization with abnormal organisms and selects resistant strains that may persist in the environment. Immunosuppressive therapy is used in a variety of conditions such as post-transplant, cancer, chemo therapy, etc. its high risk factors include the use of high doses of prednisone, hyperglycemia, granulocytopenia and renal failure. (Allen et al' 1973). Most of the bacteria involved in the Nosocomial infections are identified as follows: Acinetobacter calcoaceticus, Aeromanas hydrophila Alcaligenes species, Bacillus species, Brucella species, Campylobacter fetus, Corynebacterium and Proprionibacterium species, Dermatophilus Congolensis, Eikenella corrodes, Flavobacterium species, Homophiles species, Lactobacillus species, Listeria monocytogenes, Moraxella species, Mycoplasma pneumonia, Neisseria species, Plesiomonas shigelloids, Staphylococcus aureus, Streptococcus species vibrio cholera etc.

Several observations suggest that there are still gap in our knowledge of the pathobiology of sepsis. For instance, cultures remain negative in 30% of patients with sepsis. Negative cultures are often attributed to prior use of antibiotics, (Bone et al 1996), yet it is possible that there still exist some invisible" bacteria responsible for sepsis that evade current culture techniques.

Therefore, the objective of this study is the isolation identification of different types of bacteria prevalent on environmental, labour wards responsible for Nosocomial infections.

MATERIALS AND METHOD

Samples were collected from the floor, bed rails, wall, mattresses, malting touch and baby beds, in the labour wards of the named hospitals , using a sterilized swab stick, moistened with peptone water. The air was sampled by exposing the prepared culture plates for 24 hours. The following preparations were used. 50ml blood agar, 50ml chocolate agar, 50ml Macconkey agar and peptone water. All the media were sterilized in the autoclave at 121^0c for 15 minutes. Petridishes, pipettes and other glassware's were sterilized at 160^0c for 1 hour, using hot air oven. Samples were inoculated on already prepared agar plates, using the streaking method and incubated at 37^0c for 24 hours. After incubation, the colonies were observed for visible growth. Colonial/cultural characterization of isolates such as size, shape, colour, elevation, surface and consistency were recorded.

IDENTIFICATION OF BACTERIA: various colonies were gram stained and observed, basically for four steps each, with tap water rinse after each step. The smear was air-dried and gently heat-fixed. The slide was flooded with

crystal violet and gram iodine (brown), each for 1minute and was washed off with tap water. The gram iodine was also carefully decolourized with alcohol, then washed off with tap water and flooded with safranin (red) for 30 seconds, then washed off with tap water, it was allowed to air-dry and viewed under light microscope using x100 objective and x10 eye piece. It was observed that gram positive organisms retained the purple colour while the gram negative organisms lost the purple colour. Biochemical reactions were carried out in the isolates as follows:

Catalase test: 3% hydrogen peroxide solution (H_2O_2) was pured into a test tube and a sterile glass rod was used to remove several colonies of the test organisms immersed in the H_20_2. Observation of immediate bubbles was recorded as positive Catalase while absence of bubbles was recorded as negative Catalase.

Oxidase test: 2 drops of freshly prepared oxidize reagent was added to a filter paper placed in a clean petridish. A piece of glass rod was used to remove colonies of the test organisms, and smeared it on the filter paper. The observation of a blue-purple colour was recorded as positive oxidase reaction while the absence of such colour was recorded as negative oxidase reaction.

Citrate test: slops of the medium were prepared in bijon bottles and sterile straight wire was used. Saline suspension of the test organisms was streaked and the slopes incubated at 37^0c for 48 hours. A bright blue colour, when observed, was recorded as positive and the absence of such colour was recorded as negative result.

Indole test: Test organisms were inoculated and incubated at 35-38^0c for up to 48 hours. Them 0.5ml of kovac's reagent was added, to test for indole. Red colour in the surface layer was observed within 10minutes as positive result while absence of surface layer colour was recorded as negative result.

Ehrilich's reagent in the overnight peptone water (broth) culture (24-48 hours): 1ml of xylol was added, 0.5ml of indole reagent was layered gently and examined for 5 seconds. A pink colour indicates the presence of indole.

RESULTS

The result of the research carried out in General hospital umuguma and Umezuruike hospital, all in Owerri, Imo State, using the same samples obtained from the floor, wall, bed rails, baby beds, malting touch and mattress indicated the presence of seven predominant bacteria on the name surfaces in the labour wards as presented below:

NB: the symbols used were defined at the end of the results.

Similarly, the result presented in table 1.2 above shows that Escherichia coli was identified on the surfaces of malting touch, Floor and Bed rails in the labour ward of Umezuruike hospital and also on baby beds in the labour ward of General hospital Umuguma. This was identified using the appropriate reagents as recorded in the table.

Table 1.1 ***Staphylococcus aureus***

Colony code	Gram reaction	Catalase	Coagulase	Oxidize	Citrate	Intdole	Motility	Lactose	Glucose	Sucrose	Manitol	Probabale organism
UMT	Gram positive, cocci in clusters.	tve	tve	ND	ND	ND	ND	A	A	G	A	Staphylococcus aureus
UBB	Gram positive, cocci in clusters	tve	tve	ND	ND	ND	ND	A	A	G	A	Staphylococcus aureus
UA	Gran positive, cocci in clusters.	tve	tve	ND	ND	ND	ND	A	A	G	A	Staphylococcus aureus
GBR	Gram positive, cocci in clusters	tve	tve	ND	ND	ND	ND	A	A	G	A	Staphylococcus aureus
GF	Gram positive, cocci in clusters	tve	tve	ND	ND	ND	ND	A	A	G	A	Staphylococcus aureus
GW	Gram positive, cocci in clusters	tve	tve	ND	ND	ND	ND	A	A	G	A	Staphylococcus aureus
GA	Gram positive, cocci in clusters	tve	tve	ND	ND	ND	ND	A	A	G	A	Staphylococcus aureus

In table 1.1 above, the presence of *staphylococcus aureus* were identified on all the named surfaces in the above mentioned hospitals, using the same samples and appropriate reagents.

Table 1.2: Escherichia coli

Colony code	Gram reaction	Catalase	Coagulase	Oxidize	Citrate	Intdole	Motility	Lactose	Glucose	Sucrose	Manitol	Probabale organism
GBB	Gram negative rod	tve	ND	-ve	Tve	AG	AG	AG	AG	AG	A	*Escherichia Coli*
UMT	Gram negative rod	tve	ND	-ve	tve	AG	AG	AG	AG	AG	A	*Escherichia Coli*
UF	Gram negative rod	tve	ND	-ve	tve	AG	AG	AG	AG	AG	A	*Escherichia Coli*
UBR	Gram negative rod	tve	ND	-ve	tve	AG	AG	AG	AG	AG	A	*Escherichia Coli*

Table 1.3 Bacillus species

Colony code	Gram reaction	Catalase	Coagulase	Oxidize	Citrate	Intdole	Motility	Lactose	Glucose	Sucrose	Manitol	Probable organism
UW	Gram Negative rod	tve	ND	ND	-ve	ND	+ve	A	A	A	A	Bacillus species
UBB	Gram negative in singles	tve	ND	ND	-ve	ND	+ve	A	A	A	A	Bacillus species
UA	Gram negative rod in chains	tve	ND	ND	-ve	ND	+ve	A	A	A	A	Bacillus species
GMT	Gram Negative rod in long chains	tve	ND	ND	-ve	ND	+ve	A	A	A	A	Bacillus species

Bacillus species was also identified on the malting touch in the labour ward of General hospital Umuguma as well as on the surfaces of baby beds, wall and air in the Umezuruike hospital lablur ward. This was identified using the named reagents as presented in table 1.3, above

Table 1.4 klebsiela acrogens

Colony code	Gram reaction	Catalase	Coagulase	Oxidize	Citrate	Intdole	Motility	Lactose	Glucose	Sucrose	Manitol	Probable organism
UF	Gram Negative rod	+ve	ND	-ve	+ve	-ve	AG	AG	AG	AG	A	Klebsiela acrogens
GA	Gram Negative rod	+ve	ND	-ve	+ve	-ve	AG	AG	AG	AG	A	Klebsiela acrogens
GF	Gram negative rod	+ve	ND	-ve	+ve	-ve	AG	AG	AG	AG	A	Klebsiela acrogens

In table 1.4 above, the result shows that klebsiela acrogens was identified on the floor of Umezuruike hospital labour ward and also on the floor and in the air of General hospital Umuguma labour ward, using the appropriate reagents named in table 1.4.

Table 1.5 streptococcus faecalis

Colony code	Gram reaction	Catalase	Coagulase	Oxidize	Citrate	Intdole	Motility	Lactose	Glucose	Sucrose	Manitol	Probable organism
GMT	Gram positive, cocci in short chains.	-ve	-ve	ND	ND	ND	ND	A	A	ND	AG	Streptococcus faecalis
GM	Gram positive, cocci in short chains.	-ve	-ve	ND	ND	ND	ND	A	A	ND	AG	Streptococcus faecalis
UM	Gram positive, cocci in short chains.	-ve	-ve	ND	ND	ND	ND	A	A	ND	AG	Streptococcus faecalis

It was also observed in table 1.5 above that streptococcus faecalis was identified on the surfaces of malting touch and mattress in the labor ward of General hospitals Umuguma and also on the surface of the mattress in Umuzuruike hospital labour wards, using the appropriate named reagents in table 1.5 above.

Table 1.6: Micrococcus species

Colony code	Gram reaction	Catalase	Coagulase	Oxidize	Citrate	Intdole	Motility	Lactose	Glucose	Sucrose	Manitol	Probable organism
GF	Gram Negative, cocci in tetrads	+ve	-ve	ND	ND	ND	ND	-ve	A	-ve	-ve	Micrococcus species

Micrococcus specie was identified on the surface of the floor only in general hospital Umuguma labour ward as presented in table 1.6 above, using the appropriate named reagents as shown in the table.

Table 1.7: Pseudomonas aeruginosa

Colony code	Gram reaction	Catalase	Coagulase	Oxidize	Citrate	Intdole	Motility	Lactose	Glucose	Sucrose	Manitol	Probable organism
GBR	Gram negative rod	-ve	ND	+ve	+ve	-ve	+ve	-ve	A	-ve	AG	Pseudomonas aeruginosa
GM	Gram negative rod	-ve	ND	+ve	+ve	-ve	+ve	-ve	A	-ve	AG	Pseudomonas aeruginosa
UM_1	Gram negative rod	-ve	ND	+ve	+ve	-ve	+ve	-ve	A	-ve	AG	Pseudomonas aeruginosa
UM_2	Gram negative rod	-ve	ND	+ve	+ve	-ve	+ve	-ve	A	-ve	AG	Pseudomonas aeruginosa

Similarly, Pseudomonas aeruginosa was identified on the surface bed rails, mattress and in the air of General hospital Umuguma labour ward and also on the mattress of Umezuruike hospital labour wards; using the appropriate named reagents as shown in table 1.7 above.

DEFINITION OF SYMBOLS USED IN THE TABLES

UF = Umuezuruike hospital floor

UW = Umuezuruike hospital wall

UBB = Umuezuruike hospital baby bed

UM = Umuezuruike hospital mattress

UMT = Umuezuruike hospital malting touch

UA = Umuezuruike hospital Air

GMT = General hospital Umuguma malting touch

GBR = General hospital Umuguma bed rails

GF = General hospital Umuguma floor

GW = General hospital Umuguma wall

GBB = General hospital Umuguma baby bed

GM = General hospital Umuguma mattress

GA = General hospital Umuguma Air

ND = Not done

A = Acid

G = Gas

AG = Acid and Gas

DISCUSSION OF FINDINGS/CONCLUSION

By this research, about seven bacterial speices responsible for Nosocomial infections were identified on the named surfaces of the environmental labour wards of General hospital Umuguma and Umezuruike hospital Owerri. This was done through biochemical tests, in line with the ideas of Levy et al (2001). The presence of these organisms on the surfaces of hospitals equipments was suspected by lgnaz semmelweis as the cause of puerperal fever and death of women after delivery in the labour ward of Vienna General hospital. (Bochud, 2001). It was observed by this research that Nosocomial infections are contracted by the patients and hospital workers through contaminated equipments in hospitals. The seven bacteria identified in this regard are discussed below.

Staplylococcus aureus was identified in highest concentration, suggesting that it is the commonest cause of Nosocomial infections in the named hospitals. They can colonize interior of skin, wounds and the rectum of infected persons. They can also invade the blood and cause serious complication such as bacterenia, septic shocks, endocarditis, pneumonia, osteomylitis, arthritis, toxic shock, meningitis, etc. this was in agreement with the finding of semmelweis. (Bochud,2001) S. aureus can also cause minor skin infections such as pimples, impetigo, boils, cellulitis, carbuncles, scaled skin syndrome, abscesses, etc. (Barrett, `1971).

Bacillus species was noted for its versatility in degrading complex macro molecules. Their primary habitat is the soil and their spores are continuously dispersed by means of dust into water and on animal bodies. They are ubiquitous and cause anthrax and food poisoning.

They grow in the lungs and release exotoxins that produce toxemia with pathogenic effect to include capillary thrombosis and cardiovascular shock. These information about B. species have been discovered by Barreth (1971). According to him the effects of the B. spp. Can develop into septicemia and equally causes death pseudomonas aeryginosa was also identified as presented. The studies of Klevens, (2007) described P. aeryginosa as a bacteria that lives naturally in soil, sea and fresh water often colonize animal bodies and frequently contaminate homes and health care facilities, highly versatile, food spoilage agents, plants pathogens and genetic engineering host. Some species produce antibodies called pseudomycins, which is effective in treating fungal infections. They commonly infect compromised host with severe burns, neoplastic disease and cystic fibrosis. Its complications include pneumonia, urinary tract infection, abscesses, otittis etc.

Escharichia coli is prevalent in clinical specimens and its infection are common because it is the most aerobic and non-fastidious bacteria in the gut. Its disease are mostly transmitted exclusively among humans, as observed by Klevens, (2007) and these includes infertile diarrhea, urinary tract infections, neonatal meningitis, pneumonia, septicemia and wound infections.

Streptococcus facalis is identified as presented. It has been described as normal residents, though some are agents of disease in humans. They invade the skin and the mucus membrane of the throat, causing certain inflamatiions, marked by burning, itching papules that break and form a highly contagious yellow crust (Klevens, 2007).

Micrococcus species is the last identified bacteria which are non-motile aerobic cocci. Klevens (2007) stated that they are mostly harmless saprophytes that occur in soil and water naturally, but inhabits the skin of humans and animals, causing trases of disease.

Conclusively, these seven identified bactreria are responsible for Nosocomial infection in the environmental labour wards of General Hospital Umuaguma and Umezuruike hospital Owerri, all in Imo State, Nigeria. Among the seven identified bacteria, staphyloccus aureus was found to be highly prevalent than others, thus responsible for the common infections in the hospitals.

RECOMMENDATIONS

The following recommendation were made, depending on the findings of this research.

- Washing of hands with soap and water regularly by patients and health care workers should be practiced.

- Cuts and scraps should be cleaned and covered with bandage until they are healed.
- Sharing of personal items such as towel, razors, handkerchiefs, etc should be avoided
- Health care workers as well as patients should be educated on the importance of hand hygiene
- Regular use of aprons, gloves etc during patient care activities should be highly encouraged

With these and others put in practice, the causes of Nosocomal infection in health care outlets and hospital will certainly reduce or even totally be eliminated.

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